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 FILE 'CAPLUS' ENTERED AT 15:43:29 ON 15 JUN 1999
L1          1840 S ((SITE (3N) SPECIFIC) (5N) RECOMBIN?)
L2          1840 S L1 (3N) SITE?
L3          1840 S L1 AND L2
L4         51816 S PRIMER?
L5            28 S L3 (5N) L4
L6            13 S L5 AND PCR
L7             7 S L6  AND PY<1997
L8            15 S L1 (10N) PCR
L9            12 S L8 NOT L7
L10        23861 S TEMPLATE
L11            2 S L10 AND L9
L12           12 S L9 (5N) SITE?
L13            4 S L12 (5N) (MODIF? OR ENGINEER? OR INTRODUC? OR INSERT? OR
PLAC
                E HARTLEY JAMES/AU
L14           37 S E3 OR E11
L15           37 DUP REM L14 (0 DUPLICATES REMOVED)
L16           37 S L15
L17            1 S L1 AND
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US PAT NO: 5,912,326 [IMAGE AVAILABLE] L5: 1 of 29
DATE ISSUED: Jun. 15, 1999
TITLE: Cerebellum-derived growth factors
INVENTOR: Han Chang, Mountain View, CA
ASSIGNEE: President and Fellows of Harvard College, Cambridge, MA (U.S. corp.)
Leland S. Stanford University, Palo Alto, CA (U.S. corp.)
APPL-NO: 08/525,864
DATE FILED: Sep. 8, 1995
ART-UNIT: 165
PRIM-EXMR: Marianne P. Allen
LEGAL-REP: Giulio A. DeConti, Jr.

US PAT NO: 5,912,326 [IMAGE AVAILABLE] L5: 1 of 29

DETDESC:

DETD(28)

As used herein, a "transgenic animal" is any animal, preferably a non-human mammal, bird or an amphibian, in which one or more of the cells of the animal contain heterologous nucleic acid introduced by way of human intervention, such as by transgenic techniques well known in the art. The nucleic acid is introduced into the cell, directly or indirectly by introduction into a precursor of the cell, by way of deliberate genetic manipulation, such as by microinjection or by infection with a recombinant virus. The term genetic manipulation does not include classical cross-breeding, or in vitro fertilization, but rather is directed to the introduction of a recombinant DNA molecule. This molecule may be integrated within a chromosome, or it may be extrachromosomally replicating DNA. In the typical transgenic animals described herein, the transgene causes cells to express a recombinant form of the subject cdGF protein, e.g. either agonistic or antagonistic forms. However, transgenic animals in which the recombinant cdGF gene is silent are also contemplated, as for example, the **FLP** or **CRE recombinase** dependent **constructs** described below. The "non-human animals" of the invention include vertebrates such as rodents, non-human primates, sheep, dog, cow, chickens, amphibians, reptiles, etc. Preferred non-human animals are selected from the rodent family including rat and mouse, most preferably mouse, though transgenic amphibians, such as members of the Xenopus genus, and transgenic chickens can also provide important tools for understanding, for example, embryogenesis. The term "chimeric animal" is used herein to refer to animals in which the recombinant gene is found, or in which the recombinant is expressed in some but not all cells of the animal. The term "tissue-specific chimeric animal" indicates that the recombinant cdGF gene is present and/or expressed in some tissues but not others.

US PAT NO: 5,912,141 [IMAGE AVAILABLE] L5: 2 of 29
DATE ISSUED: Jun. 15, 1999
TITLE: Nucleic acids encoding tumor virus susceptibility genes
INVENTOR: Jurgen Brojatsch, Jamaica Pond, MA
John Naughton, Somerville, MA
John A. T. Young, Auburndale, MA
ASSIGNEE: President & Fellows of Harvard College, Cambridge, MA (U.S. corp.)
APPL-NO: 08/651,579
DATE FILED: May 22, 1996

ART-UNIT: 166
PRIM-EXMR: Lila Fe[redacted]e
ASST-EXMR: Claire M. Kaufman
LEGAL-REP: Giulio A.Lahive & Cockfield, LLP DeConti, Jr.

US PAT NO: 5,912,141 [IMAGE AVAILABLE] L5: 2 of 29

DETDESC:

DETD(23)

As used herein, a "transgenic animal" is any animal, preferably a non-human mammal, bird or an amphibian, in which one or more of the cells of the animal contain heterologous nucleic acid introduced by way of human intervention, such as by transgenic techniques well known in the art. The nucleic acid is introduced into the cell, directly or indirectly by introduction into a precursor of the cell, by way of deliberate genetic manipulation, such as by rnicroinjection or by infection with a recombinant virus. The term genetic manipulation does not include classical cross-breeding, or in vitro fertilization, but rather is directed to the introduction of a recombinant DNA molecule. This molecule may be integrated within a chromosome, or it may be extrachromosomally replicating DNA. In an exemplary transgenic animal, the transgene causes cells to express a recombinant form of a tvb protein, e.g. either agonistic or antagonistic forms. However, transgenic animals in which the recombinant tvb gene is silent are also contemplated, as for example, the **FLP** or **CRE recombinase** dependent **constructs** described below. Moreover, "transgenic animal" also includes those **recombinant** animals in which gene disruption of one or more tvb genes is caused by human intervention, including both recombination and antisense
t

L5: 4 of 29

US PAT NO: 5,885,836 [IMAGE AVAILABLE]
DATE ISSUED: Mar. 23, 1999
TITLE: FLP-mediated gene modification in mammalian cells, and compositions and cells useful therefor

INVENTOR: Geoffrey M. Wahl, San Diego, CA
Stephen V. O'Gorman, San Diego, CA

ASSIGNEE: The Salk Institute For Biological Studies, La Jolla, CA (U.S. corp.)

APPL-NO: 08/825,784
DATE FILED: Apr. 8, 1997
ART-UNIT: 163
PRIM-EXMR: Christopher S. F. Low
LEGAL-REP: Stephen E.Gray Cary Ware & Freidenrich Reiter

L5: 4 of 29

US PAT NO: 5,885,836 [IMAGE AVAILABLE]

REL-US-DATA: Continuation of Ser. No. 484,324, Jun. 7, 1995, Pat. No. 5,654,182, which is a continuation of Ser. No. 147,912, Nov. 3, 1993, which is a continuation of Ser. No. 666,252, Mar. 8, 1991, abandoned.

10/9/3 (Item 2 from file: 144)
DIALOG(R)File 144:Pascal

09380601 PASCAL No.: 91-0170979
DNA mutagenesis and recombination
JONES D H; SAKAMOTO K; VORCE R L; HOWARD B H
NIH, national cancer inst., lab. molecular biology, Bethesda MD 20892, USA
Journal: Nature (London), 1990, 344 (6268) 793-794
ISSN: 0028-0836 Availability: INIST-142; 354000004065960360/NUM
No. of Refs.: 11 ref.
Document Type: P (Serial) ; A (Analytic)
Country of Publication: United Kingdom

US PAT NO: 5,710,248 [IMAGE AVAILABLE] L4: 13 of 16
DATE ISSUED: Jan. 20, 1998
TITLE: Peptide tag for immunodetection and immunopurification
INVENTOR: Charles F. Grose, Iowa City, IA
ASSIGNEE: University of Iowa Research Foundation, Iowa City, IA (U.S. corp.)
APPL-NO: 08/681,935
DATE FILED: Jul. 29, 1996
ART-UNIT: 186
PRIM-EXMR: Frank C. Eisenschenk
ASST-EXMR: Patrick J. Nolan
LEGAL-REP: Mueting, Raasch, Gebhardt & Schwappach, P.A.

US PAT NO: 5,710,248 [IMAGE AVAILABLE] L4: 13 of 16

DRAWING DESC:

DRWD(2)

FIG. 1 diagrams a **recombination site** specific **PCR** insertional mutagenesis technique employed in this invention. In Panel A mutating primers (MP) MP15 and MP16 were used to insert 24 nucleotides (3B3.8) into the VZV gL gene immediately downstream from codon 21. The darker portions of MP15 and MP16 represent nucleotides that are complementary to the gL template (an overlap of 20 and 21 bp respectively). Panel B represents the resulting pTM1-VZVgL3B3.8 plasmid. Panel C represents PCR insertional mutagenesis where MP19 and MP20 insert 9 nucleotides (3B3.3) into the gL3B3.8 gene immediately downstream from the 3B3.8 insertion. The 3' ends of MP19 and MP20 overlap the gL3B3.8 template by 24 bp each. The 5' ends overlap the gL3B3.8 template by 9 bp and 8 bp respectively. The 3' end of MP20 and the 5' end of MP19 overlap a portion of the 3B3.8 insertion. Panel D and Panel E represent the final incorporation of the 3B3-epitope tag, designated gL3B3.11. An "*" denotes the position within the ampicillin resistance gene where the non-mutating PCR primers (P3 and P4) are located. The primers are listed in Table 2.

DETDESC:

DETD(55)

The last eight amino acid residues of 6B1 (3B3.8; QRQYGDVF, see Table 1 above) were initially inserted into VZV protein gL by a **recombination site** specific **PCR** insertional mutagenesis method. (FIG. 1, panel A). The insert was placed downstream from codon 21 of the VZV gL gene, a site known to have little effect on gL function (unpublished, see FIG. 2 for codon 21 location). The sequence of gL is provided as SEQ ID NO:17. The N-terminus sequence before the first methionine (nucleotides 1-3 on SEQ ID NO:17) is ACG TCG TAG TGA AGG GAA AAC ACA AGC GTC ATG when the terminal ATG of this sequence is nucleotides 1-3 of SEQ ID NO:17. The stop codon for the gL protein is located at position 478-480. The primers for PCR mutagenesis were also designed to incorporate a new restriction site, BstNI, into the mutated plasmid for efficient screening of positive clones. The darker portions of MP15 and MP16 (FIG. 1) represent nucleotides complementary to the DNA template (wild-type gL), an overlap of 20 and 21 bp, respectively (Table 2). The asterisk in FIG. 1 denotes the position of the non-mutating PCR primers P3 and P4, which complement an overlapping portion of the ampicillin resistance gene within pTM1 (Table 2, see Duus, et al. supra).

DETDESC:

DETD(63)

In subsequent experiments the plasmid pTM1-VZV gL3B3.8 was digested with restriction enzymes Nco I and Spe I to give two separate linear species, which served as templates for the PCR mutagenesis, with mutating primers MP19 and MP20 and with two non-mutating primers P3 and P4 (Table 2). The 3B3.3 insertional mutagenesis used paired primers MP19/P4 and MP20/P3 which produced two linear products of 2.8 kb and 3.4 kb, respectively. The insert now contained a unique MboI restriction endonuclease recognition **site**. **Recombination site** specific **PCR** insertional mutagenesis of plasmid pTM1-gL3B3.8 (6.2 kb) was performed under the same conditions as the 3B3.8 insertion. Both PCR products were co-transformed into MAX Efficient DH5a.TM. Competent Cells (Life Technologies, Gaithersberg, Md.). Ten colonies were picked and screened by PCR amplification of gL with primers P1 and P2, as described by Duus, et al. (supra). Agarose gel electrophoresis demonstrated that 8 of 10 amplified PCR clones were positive for gL sequences. Five gL positive clones were randomly chosen and digested with Mbo I. The plasmid DNA of one clone was isolated and sequenced. This plasmid was designated pTM1-gL3B3.11 and contained the 11-codon 3B3-epitope (QRQYGDVFKGD (SEQ ID NO:1)) inserted into the gL gene (FIGS. 1D, E) as confirmed by s

L15: 1 of 4

US PAT NO: 5,888,732 [IMAGE AVAILABLE]
DATE ISSUED: Mar. 30, 1999
TITLE: Recombinational cloning using engineered recombination sites

INVENTOR: James L. Hartley, Frederick, MD
Michael A. Brasch, Gaithersburg, MD

ASSIGNEE: Life Technologies, Inc., Rockville, MD (U.S. corp.)
APPL-NO: 08/663,002
DATE FILED: Jun. 7, 1996
ART-UNIT: 185
PRIM-EXMR: Nancy Degen
ASST-EXMR: William Sandals
LEGAL-REP: Sterne, Kessler, Goldstein & Fox PLLC

L15: 1 of 4

US PAT NO: 5,888,732 [IMAGE AVAILABLE]
REL-US-DATA: Continuation-in-part of Ser. No. 486,139, Jun. 7, 1995, abandoned.

DETDESC:

DETD(99)

It is important to note that as a result of the preferred embodiment being in vitro recombination reactions, non-biological molecules such as PCR products can be manipulated via the present recombinational cloning method. In one example, it is possible to clone linear molecules into circular vectors. There are a number of applications for the present invention. These uses include, but are not limited to, changing vectors, apposing promoters with genes, constructing genes for fusion proteins, changing copy number, changing replicons, cloning into phages, and cloning, e.g, **PCR** products (with an attB **site** at one end and a **loxP site** at the other end), genomic DNAs, and cDNAs.

L15: 2 of 4

S PAT NO: 5,834,202 [IMAGE AVAILABLE]
DATE ISSUED: Nov. 10, 1998
TITLE: Methods for the isothermal amplification of nucleic acid molecules

INVENTOR: Jeffrey I. Auerbach, Rockville, MD
ASSIGNEE: Replicon, Inc., Rockville, MD (U.S. corp.)
APPL-NO: 08/906,491
DATE FILED: Aug. 5, 1997
ART-UNIT: 164
PRIM-EXMR: Kenneth R. Horlick
LEGAL-REP: Jeffrey I. Auerbach

L15: 2 of 4

US PAT NO: 5,834,202 [IMAGE AVAILABLE]
REL-US-DATA: Continuation-in-part of Ser. No. 595,226, Feb. 1, 1996, Pat. No. 5,733,733, Mar. 31, 1998, which is a continuation-in-part of Ser. No. 533,852, Sep. 26, 1995, Pat. No. 5,614,389, Mar. 25, 1997, which is a continuation-in-part of Ser. No. 383,327, Feb. 3, 1995, Pat. No. 5,591,609, Jan. 7, 1997, which is a continuation-in-part of Ser. No. 933,945, Aug. 24, 1992, abandoned, which is a continuation-in-part of Ser. No. 924,643, Aug. 4, 1992, abandoned.

DETDESC:

DETD(240)

The **PCR** amplification thus yields linear double-stranded molecules having **LOX sites** on each terminus. The molecule is circularized u

? t s13/9/7, 10, 11, 12, 14, 26, 30, 33, 36 45

13/9/7 (Item 7 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

08961433 97082504
. Intergenic Flip Flop, a method for systematic gene disruption and cloning in yeast.

Mallet L; Jacquet M
Institut de Genetique et Microbiologie, URA1354 du CNRS, Universite Paris-Sud, Orsay, France.
Yeast (ENGLAND) Oct %%%1996%%%, 12 (13) p1351-7, ISSN 0749-503X Journal Code: YEA
Languages: ENGLISH
Document type: JOURNAL ARTICLE
JOURNAL ANNOUNCEMENT: 9705
Subfile: INDEX MEDICUS
We have developed a strategy named Intergenic Flip Flop which, for each gene, allows us to produce in one experiment both a disrupting cassette and a plasmid for gap repair. The same method can also be used to insert a reporter gene downstream from the promoter. This approach extends the polymerase chain reaction (PCR)-based strategy proposed by Maftahi et al. 1996. Our method consists of PCR amplification of the two flanking intergenic regions of the open reading frame (ORF) of interest, using two sets of oligonucleotides. Each PCR product is flanked by two short defined nucleotidic sequences with a unique restriction site, allowing subsequent hybridization between them. The association of the two amplimers by the complementary sequences either in the same orientation as in genomic DNA or in the opposite orientation, allows the generation, after PCR, of two distinct cassettes which can be cloned into suitable vectors. When the amplimer in the head-to-tail orientation is cloned in a vector containing a selective marker for yeast such as G418 resistance, it provides a disrupting cassette after cleavage at the unique %%%restriction%%% %%%site%%% %%%introduced%%% by the %%%PCR%%% between the two intergenic
amplimers. The amplimer with a direct orientation cloned into a yeast vector, after cleavage at the unique restriction site between the intergenic regions, permits cloning by gap repair of the gene of interest in yeast. Finally, a reporter gene can be inserted in the same plasmid. We report here the successful application of this strategy to an ORF of chromosome XIV of Saccharomyces cerevisiae: N1216.
Descriptors: *Cloning, Molecular--Methods--MT; *DNA Repair--Genetics--GE; *Mutagenesis, Insertional--Methods--MT; *Polymerase Chain Reaction--Methods --MT; *Saccharomyces cerevisiae--Genetics--GE; DNA Primers; Genes, Reporter
; Genetic Vectors--Genetics--GE; Genome, Fungal; Nucleic Acid Hybridization ; Open Reading Frames; Promoter Regions (Genetics)--Genetics--GE; Restriction Mapping; Transformation, Genetic
CAS Registry No.: 0 (DNA Primers); 0 (Genetic Vectors)

13/9/10 (Item 10 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
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08902073 97119275
Rapid detection of the Fc gamma RIIA-H/R 131 ligand-binding polymorphism using an allele-specific restriction enzyme digestion (ASRED).
Jiang XM; Arepally G; Poncz M; McKenzie SE
Division of Hematology, Children's Hospital of Philadelphia, PA, USA.
J Immunol Methods (NETHERLANDS) Nov 29 %%%1996%%%, 199 (1) p55-9,
ISSN 0022-1759 Journal Code: IFE
Contract/Grant No.: RO1 HL 54749, HL, NHLBI; R0I DK 16691, DK, NIDDK
Languages: ENGLISH
Document type: JOURNAL ARTICLE
JOURNAL ANNOUNCEMENT: 9703
Subfile: INDEX MEDICUS
A polymorphism of the gene for Fc gamma RIIA, arginine (R) or histidine (H) at position 131, alters the ability of the receptor to bind certain IgG subclasses. Identification of the Fc gamma RIIA-H/R 131 genotype has assumed increasing importance in disorders of host defense, immunohematologic diseases and systemic autoimmune disorders. We report a
new method for determination of this genotype in which an allele-specific %%%restriction%%% enzyme %%%site%%% is %%%introduced%%% into an Fc gamma
RIIA %%%PCR%%% product from genomic DNA, and polymorphism assignment is
determined by %%%restriction%%% enzyme digestion followed by agarose gel
electrophoresis. This method is more rapid, more reliable and less expensive than currently available methods.
Tags: Human; Support, Non-U.S. Gov't; Support, U.S. Gov't, P.H.S.
Descriptors: *Arginine--Metabolism--ME; *DNA Restriction Enzymes --Metabolism--ME; *Histidine--Metabolism--ME; *Polymorphism (Genetics) --Immunology--IM; *Receptors, IgG--Genetics--GE; *Receptors, IgG --Metabolism--ME; Alleles; Arginine--Immunology--IM; DNA Restriction Enzymes--Genetics--GE; Histidine--Immunology--IM; Ligands; Receptors, IgG --Analysis--AN
CAS Registry No.: 0 (Ligands); 0 (Receptors, IgG); 7004-12-8 (Arginine); 7006-35-1 (Histidine)
Enzyme No.: EC 3.1.21 (DNA Restriction Enzymes)

13/9/11 (Item 11 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
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08901630 97015957
Rapid detection of point mutations of the Neisseria gonorrhoeae gyrA gene associated with decreased susceptibilities to quinolones.
Deguchi T; Yasuda M; Nakano M; Ozeki S; Ezaki T; Maeda S; Saito I; Kawada Y
Department of Urology, Gifu University School of Medicine, Japan.
J Clin Microbiol (UNITED STATES) Sep %%%1996%%%, 34 (9) p2255-8, ISSN 0095-1137 Journal Code: HSH
Languages: ENGLISH
Document type: JOURNAL ARTICLE
JOURNAL ANNOUNCEMENT: 9703
Subfile: INDEX MEDICUS
Mutations in the gyrA gene resulting in amino acid changes at Ser-91 and Asp-95 are significantly associated with decreased susceptibilities to quinolones in Neisseria gonorrhoeae. To detect these mutations, we developed a rapid and simple assay based on amplification of the region of the gyrA gene containing the mutation sites by PCR and digestion of the PCR product with a restriction enzyme. A naturally occurring HinfI restriction site was present in the region containing the Ser-91 codon, and an artificial HinfI restriction site was created in the region containing the Asp-95 codon by the method of %%%primer%%%-specified %%%restriction%%% %%%site%%% %%%modification%%%. The mutations generating alterations at
Ser-91 and Asp-95 were detected as restriction fragment length polymorphisms of the PCR products digested with HinfI. Fifty-five clinical strains of N. gonorrhoeae were examined for mutations in the gyrA gene by this method. Mutations at Ser-91 and/or Asp-95 were detected in all the 31 strains in which the mutations had been confirmed by DNA sequencing. Our method allows simultaneous testing of a large number of strains and provides results within 8 h. This rapid and simple assay could be a useful screening device for genetic alterations associated with decreased susceptibilities to quinolones in N. gonorrhoeae and could facilitate epidemiological studies on clinical isolates of N. gonorrhoeae with decreased susceptibilities to quinolones.
Descriptors: *Drug Resistance, Microbial--Genetics--GE; *DNA Topoisomerase (ATP-Hydrolysing)--Genetics--GE; *Genes, Bacterial; *Neisseria gonorrhoeae--Genetics--GE; *Quinolones--Pharmacology--PD; Base Sequence; Molecular Sequence Data; Neisseria gonorrhoeae--Metabolism--ME;
Point Mutation; Sequence Analysis, DNA
CAS Registry No.: 0 (Quinolones)
Enzyme No.: EC 5.99.1.- (gyrA protein); EC 5.99.1.3 (DNA Topoisomerase (ATP-Hydrolysing))

13/9/12 (Item 12 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 1999 Dialog Corporation. All rts. reserv.

08878060 97018061
K-ras point mutations are rare events in premalignant forms of Barrett's oesophagus.
Trautmann B; Wittekind C; Strobel D; Meixner H; Keymling J; Gossner L; Ell C; Hahn EG
Department of Medicine I, University of Erlangen-Nuremberg, Germany.
Eur J Gastroenterol Hepatol (ENGLAND) Aug %%%1996%%%, 8 (8) p799-804,
ISSN 0954-691X Journal Code: B9X
Languages: ENGLISH
Document type: JOURNAL ARTICLE
JOURNAL ANNOUNCEMENT: 9703
Subfile: INDEX MEDICUS
OBJECTIVE: In Barrett's adenocarcinomas, in contrast to squamous oesophageal carcinomas, K-ras point mutations are thought to be a frequent event. The frequency of K-ras point mutations in premalignant forms of Barrett's oesophagus (metaplasia, dysplasia) leading to adenocarcinoma with increased risk is currently not known. To establish the frequency of K-ras mutations in premalignant forms of Barrett's oesophagus, we investigated oesophageal biopsy specimens with Barrett's metaplastic and dysplastic epithelium for point mutations in the K-ras gene/codons 12, 13. DESIGN: A total of 412 biopsies from patients with Barrett's oesophagus were histologically classified into biopsies with metaplasia (n = 252), dysplasia (n = 105) and adenocarcinoma (n = 11), as well as biopsies distant from disease (normal, n = 37 and hyperplastic squamous epithelium, n = 7). METHODS: DNA from biopsy specimens was amplified by %%%polymerase%%% %%%chain%%% %%%reaction%%% (%%%PCR%%%) with a
%%%modified%%% %%%primer%%% for generating a %%%restriction%%% %%%site%%%
in the case of wild type in codon 12. Wild-type or point mutations in the K-ras gene/codons 12, 13 were detected by restriction fragment length analysis of the PCR products. RESULTS: Point mutations in K-ras/codon 12 were found in 9 biopsies (n = 1 in metaplasia, n = 4 in dysplasias, n = 4 in adenocarcinomas). All the other biopsies showed the wild type of K-ras/codon 12. No K-ras/codon 13 mutation (GGCgly-->GACasp) was observed.
CONCLUSION: Mutations in K-ras/codon 12 were rarely found in premalignant forms of Barrett's oesophagus. Whereas the screening for K-ras point mutations in metaplastic sites of Barrett's epithelium seems not to be of practical value, the screening for mutations in dysplastic lesions might be helpful to estimate the individual risk for progression of Barrett's epithelium to adenocarcinoma. A further evaluation in larger numbers of patients is needed.
Tags: Human; Support, Non-U.S. Gov't

Descriptors: *Barrett Esophagus--Genetics--GE; *Genes, ras--Genetics--GE; *Point Mutation; *Precancerous Conditions--Genetics--GE; Adenocarcinoma --Genetics--GE; Adenocarcinoma--Pathology--PA; Barrett Esophagus --Pathology--PA; Biopsy; Disease Progression; Esophageal Neoplasms --Genetics--GE; Esophageal Neoplasms--Pathology--PA; Esophagus--Pathology --PA; Metaplasia; Polymerase Chain Reaction

13/9/14 (Item 14 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
(c) format only 1999 Dialog Corporation. All rts. reserv.

08839822 96422842
Detection of the plasma cholinesterase K variant by %%%PCR%%% using an amplification-%%%created%%% %%%restriction%%% %%%site%%%.
Jensen FS; Nielsen LR; Schwartz M
Department of Anaesthesia, National University Hospital, Copenhagen, Denmark.
Hum Hered (SWITZERLAND) Jan-Feb %%%1996%%%, 46 (1) p26-31, ISSN 0001-5652 Journal Code: GE9
Languages: ENGLISH
Document type: JOURNAL ARTICLE
JOURNAL ANNOUNCEMENT: 9702
Subfile: INDEX MEDICUS
Ten individuals registered at the Danish Cholinesterase Research Unit were examined at the DNA level for the presence of the K allele of plasma cholinesterase, using amplification-created restriction sites (ACRSs). A further nine members of a family registered at the unit were tested for mutations of the K and atypical variants. The frequency of the K allele was calculated from examination of normal material from 25 individuals, representing 50 random alleles. The results show that the ACRS method successfully demonstrates the presence of the K variant, whose frequency in the Danish population was found to be 0.18. We conclude that this technique is a reliable and rapid non-radioactive diagnostic assay for detecting the plasma cholinesterase K variant.
Tags: Female; Human; Male
Descriptors: *Cholinesterases--Genetics--GE; Alleles; Base Sequence; DNA Primers; Genotype; Molecular Sequence Data; Pedigree; Polymerase Chain Reaction; Restriction Mapping; Variation (Genetics)
CAS Registry No.: 0 (DNA Primers)
Enzyme No.: EC 3.1.1.8 (Cholinesterases)

13/9/26 (Item 26 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
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08742888 96292642
Molecular diagnosis of exocrine pancreatic cancer using a percutaneous technique.
Evans DB; Frazier ML; Charnsangavej C; Katz RL; Larry L; Abbruzzese JL
Department of Surgical Oncology, University of Texas M.D. Anderson Cancer Center, Houston 77030, USA.
Ann Surg Oncol (UNITED STATES) May %%%1996%%%, 3 (3) p241-6, ISSN 1068-9265 Journal Code: B9R
Contract/Grant No.: CA 16672, CA, NCI
Languages: ENGLISH
Document type: CLINICAL TRIAL; JOURNAL ARTICLE
JOURNAL ANNOUNCEMENT: 9611
Subfile: INDEX MEDICUS
BACKGROUND: The K-ras oncogene is activated by point mutations at codon 12 in most patients with exocrine pancreatic cancer. Mutant-enriched polymerase chain reaction (PCR) amplification can enhance the detection of mutated K-ras. This technique was applied to patients undergoing percutaneous fine-needle aspiration (FNA) biopsy of suspect pancreatic lesions. METHODS: Twenty-five patients underwent percutaneous FNA of the pancreas for cytologic and molecular analysis. After preparing cytologic smears, the 22-gauge needle and syringe used for FNA were rinsed in RPMI-1640. The specimen was centrifuged, and DNA was extracted from the supernatant and subjected to mutant-enriched %%%PCR%%% using appropriate mismatched %%%primers%%% that %%%introduce%%% a BstNI %%%restriction%%% endonuclease clevage %%%site%%% at codon 12 of wild-type, but not mutant, K-ras. After digestion with BstNI, the DNA was reamplified. To increase assay sensitivity, the final five PCR cycles were completed incorporating 5 microCi of (alpha-32P)dCTP. The DNA was then redigested and subjected to gel electrophoresis and autoradiography. RESULTS: The median amount of DNA retrieved per specimen was 3.33 micrograms. Mutant K-ras was detected as a band of 143 bps; residual wild-type DNA was seen as a 114-bp fragment. Twenty-one of 25 specimens demonstrated mutated K-ras DNA. Two patients with nondiagnostic cytology results had mutated K-ras DNA; adenocarcinoma of pancreatic origin was confirmed in both cases after pancreatectomy. CONCLUSION: The molecular diagnosis of pancreatic cancer through identifications of mutations in K-ras can be readily performed on specimens obtained by percutaneous FNA. As aggressive multimodality management of this disease becomes more common, pretreatment analysis of molecular determinants may have greater clinical significance.
Tags: Human; Support, Non-U.S. Gov't; Support, U.S. Gov't, P.H.S.
Descriptors: *Pancreatic Neoplasms--Diagnosis--DI; Adenocarcinoma --Diagnosis--DI; Adenocarcinoma--Genetics--GE; Adenocarcinoma--Pathology --PA; Base Sequence; Biopsy, Needle--Methods--MT; Codon; DNA Mutational Analysis; Feasibility Studies; Genes, ras--Genetics--GE; Molecular Sequence Data; Pancreas--Pathology--PA; Pancreatic Neoplasms--Genetics--GE; Pancreatic Neoplasms--Pathology--PA; Point Mutation; Polymerase Chain Reaction--Methods--MT; Predictive Value of Tests
CAS Registry No.: 0 (Codon)

13/9/30 (Item 30 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
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08684189 96186951
%%%Creating%%% seamless junctions independent of %%%restriction%%% %%%sites%%% in %%%PCR%%% cloning.
Padgett KA; Sorge JA
Stratagene, La Jolla, CA 92037, USA.
Gene (NETHERLANDS) Feb 2 %%%1996%%%, 168 (1) p31-5, ISSN 0378-1119
Journal Code: FOP
Languages: ENGLISH
Document type: JOURNAL ARTICLE
JOURNAL ANNOUNCEMENT: 9608
Subfile: INDEX MEDICUS
A method is described for the efficient cloning of any given DNA sequence into any desired location without the limitation of naturally occurring restriction sites. The technique employs the polymerase chain reaction (PCR) combined with the capacity of the type-IIS restriction endonuclease (ENase) Eam1104I to cut outside its recognition sequence. Primers that contain the Eam1104I recognition site (5'-CTCTTC) are used to amplify the DNA fragments being manipulated. Because the ENase is inhibited by site-specific methylation in the recognition sequence, all internal Eam1104I sites present in the DNA can be protected by performing the PCR amplification in the presence of 5-methyldeoxycytosine (m5dCTP). The primer-encoded Eam1104I sites are not affected by the modified nucleotides (nt) since the newly synthesized strand does not contain any cytosine residues in the recognition sequence. In addition, the ENase's ability to cleave several bases downstream from its recognition site allows the removal of superfluous, terminal sequences from the amplified DNA fragments, resulting in 5' overhangs that are defined by the nt present within the cleavage site. Thus, the elimination of extraneous nt and the generation of unique, non-palindromic sticky ends permits the formation of seamless junctions in a directional fashion during the subsequent ligation event.
Descriptors: *Cloning, Molecular--Methods--MT; *Deoxyribonucleases, Type II Site-Specific--Metabolism--ME; *Polymerase Chain Reaction; Base Sequence ; Deoxycytidine--Analogs and Derivatives--AA; Deoxycytidine--Pharmacology --PD; DNA Primers--Chemistry--CH; Electrophoresis, Polyacrylamide Gel; Genetic Vectors--Genetics--GE; Methylation; Molecular Sequence Data
CAS Registry No.: 0 (DNA Primers); 0 (Genetic Vectors); 838-07-3 (5-methyldeoxycytidine); 951-77-9 (Deoxycytidine)
Enzyme No.: EC 3.1.21.4 (Deoxyribonucleases, Type II Site-Specific)

13/9/33 (Item 33 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
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08639162 98040678
Design of cassette baculovirus vectors for the production of therapeutic antibodies in insect cells.
Poul MA; Cerutti M; Chaabihi H; Devauchelle G; Kaczorek M; Lefranc MP
Laboratoire d'ImmunoGenetique Moleculaire, Institut de Genetique Moleculaire, UMR 9942, CNRS, Montpellier, France.
Immunotechnology (NETHERLANDS) Dec %%%1995%%%, 1 (3-4) p189-96, ISSN 1380-2933 Journal Code: CR0
Languages: ENGLISH
Document type: JOURNAL ARTICLE
JOURNAL ANNOUNCEMENT: 9803
Subfile: INDEX MEDICUS
BACKGROUND: Various systems have been described for the expression of recombinant monoclonal antibodies for therapeutical applications. Insect cells offer great advantages with respect to post-translational modifications, stability, yields and applications. OBJECTIVES: To construct plasmid cassette transfer vectors in order to express chimeric, humanized or human antibodies in insect cells using baculovirus expression system. STUDY DESIGN: Two transfer vectors, pBHuC kappa and pBHuC gamma 1, were designed. They contain a viral promoter (polyhedrin or p10 promoters, respectively), a signal peptide sequence and a human immunoglobulin light chain C kappa gene or heavy chain C gamma 1 sequence, respectively. %%%Restriction%%% %%%sites%%% have been %%%introduced%%% to allow %%%insertion%%% of rearranged variable genes, after amplification by %%%polymerase%%% %%%chain%%% %%%reaction%%% . RESULTS: Recombinant baculoviruses expressing complete immunoglobulins have been generated by a

double-recombination event between baculovirus DNA and the loaded cassette transfer vectors. CONCLUSION: Our genetic cassette approach makes this system a very flexible and convenient one for the rapid production of therapeutic monoclonal antibodies with heavy and light chains of any human isotype. Specific variable regions selected by the antibody phage display technology can be easily transferred in these vectors to obtain a complete antibody.

Tags: Animal; Human; Support, Non-U.S. Gov't

Descriptors: *Antibodies, Monoclonal--Biosynthesis--BI; *Baculoviridae; *Genetic Vectors; *Mutagenesis, Insertional--Methods--MT; Base Sequence; Chimeric Proteins--Genetics--GE; Cloning, Molecular; Enzyme-Linked Immunosorbent Assay; Immunoglobulin Variable Region--Genetics--GE; Immunoglobulins, Heavy-Chain--Genetics--GE; Immunoglobulins, Light-Chain --Genetics--GE; Jurkat Cells; Molecular Sequence Data; Recombinant Proteins --Biosynthesis--BI; Spodoptera

CAS Registry No.: 0 (Antibodies, Monoclonal); 0 (Chimeric Proteins); 0 (Genetic Vectors); 0 (Immunoglobulin Variable Region); 0 (Immunoglobulins, Heavy-Chain); 0 (Immunoglobulins, Light-Chain); 0 (Recombinant Proteins)

13/9/36 (Item 36 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
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08596866 96423318

Detection of germline mutations in the von Hippel-Lindau disease gene by the primer specified restriction map modification method.

Kishida T; Chen F; Lerman MI; Zbar B

Laboratory of Immunobiology, National Cancer Institute-Frederick Cancer Research and Development Center, MD 21702-1201, USA.

J Med Genet (ENGLAND) Dec %%%1995%%%, 32 (12) p938-41, ISSN 0022-2593

Journal Code: J1F

Languages: ENGLISH

Document type: JOURNAL ARTICLE

JOURNAL ANNOUNCEMENT: 9702

Subfile: INDEX MEDICUS

Von Hippel-Lindau disease (VHL) is an inherited disorder characterised by a predisposition to develop tumours in the eyes, central nervous system, kidneys, and adrenal glands. Recently the VHL gene was cloned and shown to be mutated in 75% of US and Canadian VHL families. To develop simple, rapid methods for the detection of mutations found in large numbers of affected people, we designed based on the %%%primer%%% specified %%%restriction%%% %%%site%%% %%%modification%%% method. These tests have proved useful in identifying asymptomatic mutated VHL gene carriers who have the nt 505 T to C mutation or the nt 686 T to C mutation. Together with an MspI digestion test which can detect a mutation hot spot in codon 238, polymerase chain reaction/restriction endonuclease based tests can now detect VHL mutations in more than 50% of VHL type 2 families.

Tags: Female; Human; Male

Descriptors: *Germ-Line Mutation--Genetics--GE; *Hippel-Lindau Disease --Genetics--GE; Base Sequence; DNA Primers; Molecular Sequence Data; Pedigree; Restriction Mapping

CAS Registry No.: 0 (DNA Primers)

13/9/45 (Item 45 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
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08395203 95380961

Tumour diagnosis by PCR-based detection of tumour cells.

Nollau P; Jung R; Neumaier M; Wagener C

Abteilung fur Klinische Chemie, Medizinische Klinik, Universitetskrankenhaus Eppendorf, Hamburg, Germany.

Scand J Clin Lab Invest Suppl (NORWAY) %%%1995%%%, 221 p116-21, ISSN 0085-591X Journal Code: UCR

Languages: ENGLISH

Document type: JOURNAL ARTICLE; REVIEW; REVIEW, TUTORIAL

JOURNAL ANNOUNCEMENT: 9512

Subfile: INDEX MEDICUS

Tumour cells shed from solid primary tumours can be detected by the polymerase chain reaction (PCR) based on the selective amplification of mutated tumour genes or of genes expressed in a tissue specific manner. When tumour specific alterations are amplified, few tumour cells can be detected in excess of normal cells derived from the same tissue. Thus, malignant cells can be detected specifically in pancreatic juice, stool, urine, and sputum. Here we describe the adaptation of the mutant enriched %%%PCR%%% in conjunction with the %%%introduction%%% of artificial %%%primer%%% mediated %%%restriction%%% %%%sites%%% to the selective amplification of mutant K-ras genes in stool samples from patients with colorectal carcinomas. In reconstitution experiments, down to 10 colorectal carcinoma cells could be detected in 100 mg of stool. For the diagnosis of micrometastatic disease, a sensitive and specific technique was established based on the reverse transcription of mRNA specific for the carcinoembryonic antigen followed by the amplification of the cDNA (RT-PCR). Attempts to establish a specific RT-PCR for cytokeratin-18 failed because of the existence of at least one processed pseudogene. (28 Refs.)

Tags: Human

Descriptors: *Neoplasms--Diagnosis--DI; *Polymerase Chain Reaction --Methods--MT; *Tumor Markers, Biological--Diagnostic Use--DU; Neoplasms --Genetics--GE; RNA, Messenger--Analysis--AN; Tumor Markers, Biological --Genetics--GE

CAS Registry No.: 0 (RNA, Messenger); 0 (Tumor Markers, Biological)

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FTSNET 0.450 Hrs.
$86.80 Estimated cost this search
$86.91 Estimated total session cost 6.512 DialUnits
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